Right ventricular adaptation to chronic abnormal loading

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Systematic Review of Volume Load-Induced Right Ventricular Dysfunction in Animal Models: A Translational Gap in Congenital Heart Disease

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ABSTRACT

Objective: To systematically review and combine available data regarding volume load-induced RV dysfunction in animal models.

Background: The improved treatment of patients with congenital heart diseases has led to a growing group of survivors living with residual abnormalities. The most prevalent is increased right ventricular (RV) volume load that eventually induces RV dysfunction. It is unclear how to recognise and treat volume load-induced RV dysfunction.

Methods and results: MEDLINE and EMBASE were searched using a pre-specified search strategy. 18 articles met the inclusion criteria. Ventricular volume data, variables of functional adaptation and ventricular remodelling were extracted and combined using random-effects meta-analysis (Hedges' g (HG). We identified two models of RV volume overload in five species: pulmonary regurgitation and aorto-caval shunt. Meta-analysis demonstrated a time-dependent decrease in cardiac output (HG 1.1, CI 0.3;1.8). RV end-diastolic pressure increased in both models (HG 3.4, CI 2.1;4.7), whereas preload-recruitable stroke work was decreased only in the pulmonary regurgitation model (HG -1.8, CI -2.9;-0.9)). RV weight consistently increased (HG 4.0, CI 2.9;5.1) and late RV fibrosis was observed in both models. Various variables describing RV remodelling were reported inconsistently, hampering meta-analyses.

Conclusions: This systematic review describes the adaptation pattern of volume overload-induced RV remodelling in two models. Both models consistently induced increased RV EDP and RV weight and revealed a pattern of late fibrosis, which may be a potential target for therapy. The lack of structured analyses highlights the clearly existing gaps in our understanding of the volume loaded RV in animal models.

INTRODUCTION

The improved survival of children with congenital heart disease has led to a new generation of adolescents at risk for chronic heart diseases.[1,2] Due to longstanding residual lesions after correction or palliation of their cardiac defect, these adolescents are prone to develop heart failure. In the early days of corrective surgery, several of these residual lesions were erroneously perceived as benign.[3] The most prevalent of these is pulmonary regurgitation after correction for Tetralogy of Fallot, leading to right ventricular (RV) volume overload. Longstanding RV volume load is now known to induce RV dysfunction and failure.[4,5] At present, still many questions remain unanswered. It is unclear how to recognise reversible versus irreversible RV failure of a volume loaded RV and how to prevent or support a failing volume loaded RV. To answer these questions, experimental and clinical data are needed that allow the interrogation of the functional, cellular and molecular pathophysiology of RV adaptation to volume overload.[6]

To study RV adaptation to volume load, several animal models mimicking specific types of volume overload have been designed. Ideally, such models should be able to induce a standardised volume overload and should yield valuable data that allow the creation of a spatiotemporal profile of functional and cellular adaptation. However, translation of results from such animal studies to human pathophysiology is challenged by inconsistencies in model types and contradictory findings between research groups. Systematic reviews and meta-analyses of animal studies have proven very useful to contribute to a reduction and refinement of animal experiments by overcoming these challenges and by identifying and defining needs for standardization.[7,8] Therefore we aimed to systematically review and combine available experimental data from animal models and focus on the functional, cellular and molecular adaptation mechanisms involved in volume load-induced RV dysfunction.

METHODS

Literature search

We performed a systematic literature search in Medline and EMBASE on August 26th 2015. The search strategy and methodological protocol were published *a priori* on the online platform of the working group Collaborative Approach to Meta-Analysis and Review of Animal Data from Experimental Studies (CAMARADES).[9] The search strategy was composed to capture the overlapping part of the following four domains: 1) animal study; 2) RV; 3) volume load; 4) pathophysiological effects (see supplementary table 1). We used a recently published search filter for animal experiments to create the search domain "animal study".[10,11] To ensure inclusion of all juvenile animal models, we added juvenile names of animals to this search filter.

Study selection

Two researchers (G.P.L.B. and Q.A.J.H.) independently screened the primarily identified abstracts according to the following inclusion criteria: 1) original study was written in English; 2) article contained animal research; 3) RV was studied; 4) RV was subjected to >24 hours of isolated volume overload; 5) Degree of RV volume load was quantified and reported as: RV end-diastolic volume and/or area (EDV/A), RV end-systolic volume and/or area (ESV/A), cardiac output (CO) or RV stroke volume (SV); 6) a sham-operated control group was described and 7) RV adaptation was studied. Reviews and case reports were excluded. Disagreements between the reviewers were resolved by discussion leading to consensus or by consulting a third-party arbitrator (B.B.).

Data extraction

Outcome measures to be extracted were predefined in the systematic review protocol. The following methodological choices were made during the extraction phase. If separate experimental groups were sacrificed at different time points in one paper, we considered the groups as unique experiments. As statistical synthesis of scarcely reported variables would not contribute, we chose to extract variables reported in at least five experiments. Extracted data were subdivided into two groups: functional adaptation and cellular / molecular adaptation. End-diastolic volume and end-diastolic area were combined (EDV/A), since both variables represent ventricular dimensions, allowing for relative comparison. In a similar fashion, end-systolic volume and area (ESV/A) were combined. We extracted means and standard deviations of the intervention group and control group, or recalculated the standard deviations from the standard error of the mean or 95% confidence interval. The experiment-level standardised mean differences were calculated by dividing the mean difference between intervention and control groups by the pooled standard deviation, corrected for upward bias using Hedges' g.[12] This allowed for a uniform reporting of differences relative to control group values.

Data synthesis

We conducted multiple separate random-effects meta-analyses to calculate combined effect estimates. We used Cochran's Q-test and the I^2 quantity to test for between-study heterogeneity. When we found evidence for substantial heterogeneity, (Q-test p-value <0.10 or I^2 value >50%), we performed univariable meta-regression to explore the following three potential explanations for the identified between-study variety: 1) different durations of volume load (expressed as experiment time), 2) different degrees of volume overload (expressed as RV EDV/A) and 3) different model types (either shunt or regurgitation). We considered P-values <0.05 as statistically significant. We used STATA 14.0 (STATA corp., College Station, Texas, USA) for statistical analysis. We displayed the subtracted effect estimates of the in forest plots, and displayed different models of volume overload separately.

RESULTS

Studies

We identified 1220 unique citations, of which 105 fulfilled criteria for inclusion for full text review. Of these, 18 citations were included for data extraction (figure 1), combining data derived from a total of 467 animals.[13–30] From the included citations, 15 variables were reported at least five times, of which data were extracted (table 1). We combined hemodynamic data of the two included articles of Borgdorff et. al., and likewise combined the data of the articles of Ersboell et. al., Kjaergaard et. al. and Smith et. al. due to the repeated use of results involving the same animals.[15,16,19,22,29] We referred to the reference that was published first, i.e. Borgdorff 2012 and Kjaergaard 2010 respectively. Results are displayed in table 2 and correspond with the forest plots as displayed in the supplemental material. In a total of five different species (rat, mouse, pig, dog, lamb), the following two experimental models of RV volume overload were identified: the aorto-caval shunt model and the pulmonary regurgitation model. Also, five articles described a model of tricuspid regurgitation and five described a model of atrial septal defect, but none of these met the requirements for inclusion.

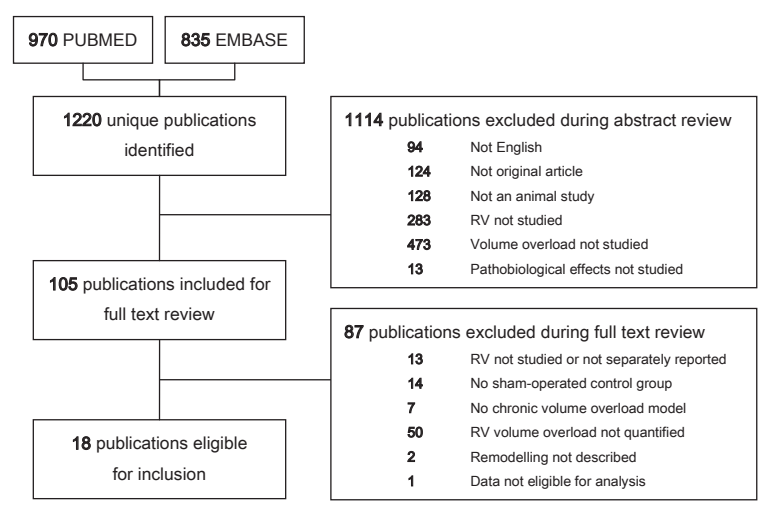

Figure 1 | Flowchart showing study selection. RV indicates right ventricle.

Table 1 | Study characteristics.

	Agger 2010(13)	Bartelds 2011(14)	Borgdorff 2012(15)	Borgdorff 2013(16)	Bove 2012(17)	Bove 2014(18)	Ersboell 2012(19)	Gray 2003(20)	Hyldebrandt 2015(21)	Kjaergaard 2010(22)	Kuehne 2003(23)	Liu 1991 chapter 1(24)	Liu 1991 chapter 2(25)	Modesti 2004(26)	Reddy 2013(27)	Ruzicka 1993(28)	Smith 2012(29)	Szabó 2006(30)	**Number of studies**	**Number of time points**
Species																				
Mouse		1													1				**2**	
Rat			1	1								1	1			1			**5**	
Sheep								1											**1**	
Dog																		1	**1**	
Pig	1				1	1	1		1	1	1			1			1		**9**	
Disease model																				
Shunt		1	1	1								1	1	1		1		1	**8**	
Regurgitation	1				1	1	1	1	1	1	1				1		1		**10**	
Developmental stage at intervention																				
Pre-pubertal	1				1	1	1	1	1	1	1						1		**9**	
Post-pubertal		1	1	1								1	1	1	1	1		1	**9**	
Hemodynamic parameters																				
RV EDV/A	1	1		1	1	1	3		1	3	1				3		1	1	**12**	**18**
RV ESV/A	1	1		1	1	1	3			3	1				3		1	1	**11**	**17**
CO	1	1	1	1	1	1	3		1		1	1	2	4		2		1	**14**	**21**
RV SV	1	1	1	1	1	1	3		1		1	1	2	4	3	2			**14**	**23**

Table 1 | Continued

	Agger 2010(13)	Bartelds 2011(14)	Borgdorff 2012(15)	Borgdorff 2013(16)	Bove 2012(17)	Bove 2014(18)	Ersboell 2012(19)	Gray 2003(20)	Hyldebrandt 2015(21)	Kjaergaard 2010(22)	Kuehne 2003(23)	Liu 1991 chapter 1(24)	Liu 1991 chapter 2(25)	Modesti 2004(26)	Reddy 2013(27)	Ruzicka 1993(28)	Smith 2012(29)	Szabó 2006(30)	**Number of studies**	**Number of time points**
PRF	1				1	1	3	1			1								**6**	**8**
RV EF	1	1	1	1	1	1	3				1			4	3				**10**	**17**
RV EDP			1	1	1	1						1	2	4	3			1	**9**	**15**
RV ESP		1			1	1					1			4				1	**6**	**9**
HR	1	1	1	1	1	1			1	3	1	1	2	4	3	2		1	**15**	**24**
RV PRSW	1		1	1		1			1									1	**6**	**6**
RV SW	1	1	1		1	1													**5**	**5**
RV dP/dt max	1			1					1		1	1	2		3				**7**	**10**
TAPSE			1			1				3							1		**4**	**6**
RV ESPVR	1		1		1	1					1							1	**6**	**6**

Data of species and disease models are presented as 1 for described in article. Data of hemodynamic variables are presented as number of time points described in study. RV indicates right ventricle. EDV/A indicates end-diastolic volume/end-diastolic area. ESV/A indicates end-systolic volume/end-systolic area. CO indicates cardiac output. SV indicates stroke volume. PRF indicates pulmonary regurgitation fraction. EF indicates ejection fraction. EDP indicates end-diastolic pressure. ESP indicates end-systolic pressure. HR indicates heart rate. PRSW indicates preload recruitable stroke work. SW indicates stroke work. dP/dt max indicates maximum rate of pressure rise. TAPSE indicates tricuspid annular plane systolic excursion. ESPVR indicates end-systolic pressure-volume relationship.

Table 2 | Hemodynamic characteristics.

Parameter	Shunt				Regurgitation				Combined			
	n	Hedges' g	95% CI	*p*	n	Hedges' g	95% CI	*p*	n	Hedges' g	95% CI	*p*	I^2(%)
RV EDV/A	220	3.12	2.16;4.08	**<0.001**	84.4	269	2.94	2.12;3.72	**<0.001**	82.8	0.45;4.28	**0.016**	84.4
RV ESV/A	181	2.64	1.84;3.44	**<0.001**	75.2	230	2.42	2.00;3.15	**<0.001**	77.9	0.01;3.46	**0.049**	84.7
RV SV	207	2.61	1.34;3.88	**<0.001**	97.7	334	2.35	1.65;3.05	**<0.001**	84.3	1.50;2.37	**<0.001**	10.9
PRF	108	6.61	4.47;8.76	**<0.001**	82.7	-	-	-	-	-	-	-	-
RV EF	168	-0.27	-0.27;0.71	0.586	88.6	223	-0.18	-0.87;0.50	0.603	83.7	-0.90;0.72	0.833	62.3
RV EDP	105	3.96	1.01;6.91	**0.008**	95.6	218	3.42	2.1;4.75	**<0.001**	91.1	1.55;4.06	**<0.001**	81.4
RV ESP	33	-0.24	-0.87;0.40	0.464	0.0	69	0.47	-0.39;1.34	0.281	68.8	-0.26;2.57	0.109	73.4
HR	221	0.23	-0.52;0.97	0.550	85.4	366	0.28	-0.18;0.73	0.232	77.2	-0.26;0.78	0.328	60.6
RV PRSW	76	-2.02	-3.08;-0.96	**<0.001**	67.2	114	-1.17	-2.35;0.00	**0.050**	86.5	-0.10;1.17	0.101	0.0
RV SW	43	0.72	-0.66;2.10	0.304	76.9	74	2.10	0.34;3.86	**0.019**	88.2	1.23;8.17	**0.008**	77.9
RV dP/dt max	137	2.21	-0.21;4.62	0.074	96.1	204	2.08	0.62;3.54	**0.005**	94.1	1.12;2.70	**<0.001**	49.3
TAPSE	75	0.00	-0.56;0.55	0.989	66.0	95	0.35	-0.60;1.29	0.473	80.7	0.89;2.94	0.314	76.2
RV ESPVR	54	-2.25	-4.30;-0.21	**0.031**	87.0	92	-1.44	-2.58;-0.29	**0.014**	81.4	-2.58;-0.29	0.224	31.6

CI indicates confidence interval. RV indicates right ventricle. EDV/A indicates end-systolic volume/end-diastolic area. ESV/A indicates end-systolic volume/end-diastolic area. SV indicates stroke volume. PRF indicates pulmonary regurgitation fraction. EF indicates ejection fraction. EDP indicates end-diastolic pressure. ESP indicates end-systolic pressure. HR indicates heart rate. PRSW indicates preload recruitable stroke work. SW indicates stroke work. dP/dt max indicates maximum rate of pressure rise. TAPSE indicates tricuspid annular plane systolic excursion. ESPVR indicates end-systolic pressure-volume relationship.

Functional RV adaptation

Both RV EDV/A and RV ESV/A were significantly increased in the two model groups compared to controls, ($p<0.001$), indicating effective induction of experimental RV volume load. CO was significantly increased in intervention groups compared to controls, in shunt models only (shunt: $p<0.001$; regurgitation: $p=0.737$, figure 2).

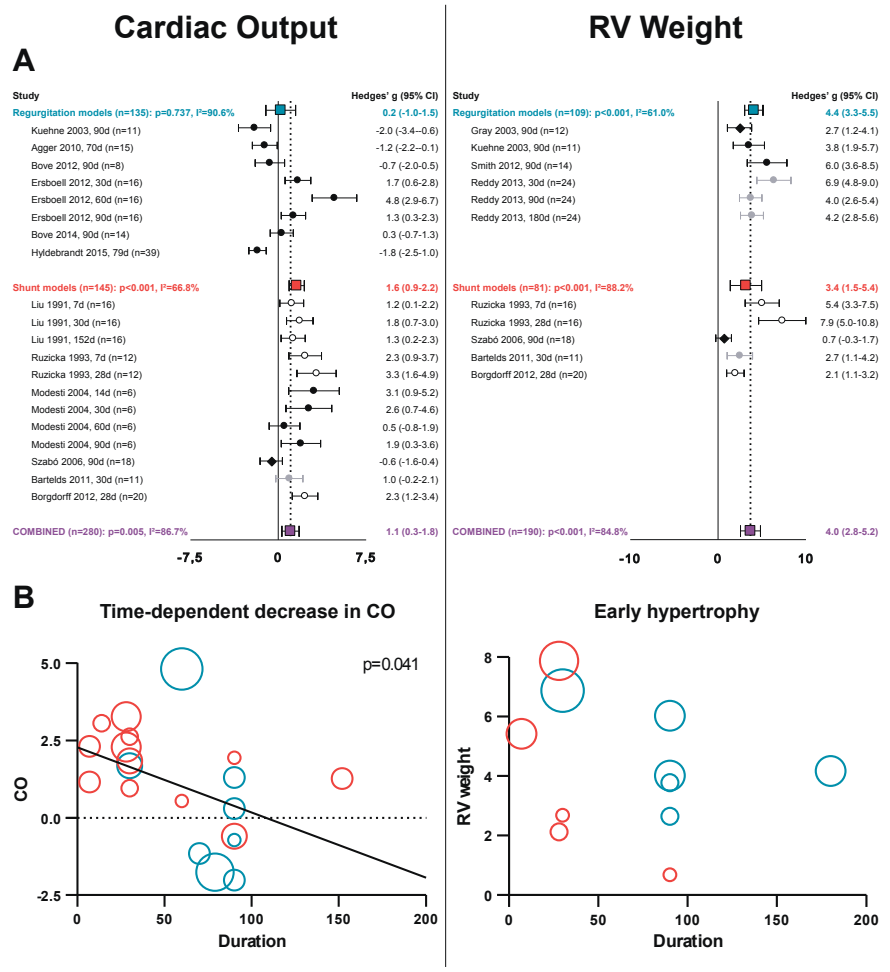

Figure 2 | A. Forest plots showing cardiac output and right ventricular (RV) weight. Data are presented as Hedges' g (95% confidence interval). CI indicates confidence interval. CO indicates cardiac output. Combined Hedges' g displayed as squares ■, pig models displayed as black dots •, mouse models displayed as grey dots •, rat models displayed as white dots ○, models with other animals displayed as diamonds ♦. Bars represent 95% confidence interval (CI). B. Bubble plots showing relations between the variables shown in A and volume load duration. Data are presented as Hedges' g. Volume load duration is presented in days. Bubble size represents study precision, calculated as standard deviation times group size. Red bubbles represent shunt models. Blue bubbles represent regurgitation models.

Pulmonary regurgitation fraction was significantly increased in all regurgitation studies (p<0.001). RV SV was significantly increased in intervention groups compared to controls, in both models (p<0.001). RV ejection fraction (EF), a parameter frequently used to assess RV failure in clinical practice, did not consistently change in either of the models. RV preload recruitable stroke work (PRSW) and RV end-systolic pressure-volume relation (ESPVR), both parameters of contractility, were significantly decreased in intervention groups compared to controls, in regurgitation models only (p<0.001 and p=0.031 respectively). RV dP/dt max was significantly increased in intervention groups compared to controls, in shunt models only (p<0.001). The RV end-diastolic pressure volume relation (EDPVR) was not consistently reported in the studies, which hampered meta-analysis. Tricuspid annular plane systolic excursion (TAPSE) did not significantly differ between intervention groups and controls. RV dP/dt min was not consistently reported. RV end-diastolic pressure (EDP), a surrogate for impaired ventricular function, was significantly increased in intervention groups compared to controls, in both models (p<0.001).

Significant heterogeneity was present in all meta-analyses (I^2 >50%), indicating that the effect sizes of the included studies varied substantially. Hence, separate univariable meta-regression analyses were performed to explore the potential role of differences in (1) durations of volume load, (2) degrees of volume load and (3) model types, in explaining this between-study heterogeneity.

Duration of volume load. Meta-regression revealed that longer duration of volume overload was significantly associated with lower CO effect estimates (p=0.041, figure 2B).

Degree of volume load. More volume load in the models (defined as higher effect estimates of EDV/A), was significantly associated with higher CO effect estimates (p=0.027), higher RV SV effect estimates (p=0.005), higher HR effect estimates (p=0.021), higher RV dP/dt max effect estimates (p=0.036) and lower RV FS effect estimates (p=0.038).

Model type. None of the parameters showed a significant association with model type, although a trend towards higher CO in shunt models was observed (p=0.054).

Ventricular remodelling

In both models, RV weight was significantly increased in intervention groups compared to controls, indicating the consistent occurrence of RV hypertrophy (p<0.001). The individual study-level effect estimates show that RV hypertrophy is independent of the duration of volume load and also present in short-duration models (figure 2b). Figure 3 lists articles reporting occurrence of fibrosis or β-myosin heavy chain/α-myosin heavy chain (β-MHC/α-MHC) ratio change. Fibrosis is consistently reported after 90 days of volume load duration or longer, whereas this is not the case at earlier time points. In contrast, the presence of β-MHC/α-MHC ratio change is independent of volume load duration. Molecular responses were described in less than five papers, precluding meta-analysis (table 3). The scarce description

of cellular adaptation reveals that plasma renin and RV renin are activated early after induction of a volume load and remain so after more than one month.[28] There is a modest activation of inductors of hypertrophy, e.g. angiotensin-II, endothelin-1, insulin-like growth factor 1 (IGF-1), regulator of calcineurin 1 (RCAN1).[14,16,26] A gene array analysis study in mice revealed a switch in transforming growth factor β (TGF-β) expression between 1 and 3 months of RV volume load.[27] Also, metabolic pathways, G protein-coupled receptors and calcium-signaling were reported to be downregulated during volume loading. Another study in mice reported downregulation of phosphor-ERK but not phospho-Akt in response to volume loading.[14] There were no studies with interfering strategies revealing the functional role of the described changes.

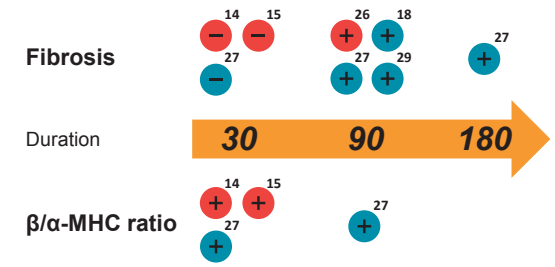

Figure 3 | Figure showing development of fibrosis and β/α-myosin heavy chain ratio (β/α-MHC ratio) changes over time. Volume load duration is presented in days. Minus sign (-) indicates that no change was detected. Plus sign (+) indicates that an increase was detected. Red dots represent shunt models. Blue dots represent regurgitation models. Numbers in superscript refer to included articles in this systematic review.

Table 3 | Molecular responses studied in experimental volume overload.

	Up	*Stable*	*Down*
Natriuretic peptides	Rat shunt 28d (15, 16) Mouse reg. 90d (27)	Mouse shunt 28d (14)	Mouse reg. 30d (27)
RAAS	Pig shunt 14,30,60,90d (26) Rat shunt 7,28d (28)		
G-protein signaling			Mouse reg. 30,90,180d (27)
Endothelin	Pig shunt 90d (26)		
Calcineurin	Rat shunt 28d (15, 16)	Mouse shunt 28d (14)	
AKT		Rat shunt 28d (15, 16) Mouse shunt 28d (14)	
ERK		Rat shunt 28d (15,16)	Mouse shunt 28d (14)
Collagen, TGF-β	Mouse reg. 90d (27)	Mouse shunt 28d (14)	Mouse reg. 30d (27)
Glucose metabolism			Mouse reg. 30d (27)
Fatty acid metabolism			Mouse reg. 90d (27)

Data are presented as upregulation (up), no change in regulation (stable) and downregulation (down). Reg. indicates regurgitation. RAAS indicates renin-angiotensin-aldosterone-system. ERK indicates extracellular signal-regulated kinase. TGF-β indicates transforming growth factor β.

DISCUSSION

To our knowledge, this is the first systematic review on RV remodelling in response to chronic volume overload in animal models. Effective volume overloading was confirmed in two experimental models: the aorto-caval shunt model and the pulmonary regurgitation model. This was associated with significant haemodynamic changes indicative of RV dysfunction. RV adaptation to volume overload was characterised by early onset of RV hypertrophy and late onset fibrosis. Studies of cellular and molecular adaptation were scarce, hampering the interpretation of molecular pathways involved in the development of RV dysfunction.

This review aims to provide an overview of RV adaptation patterns in response to volume overload. In this systematic literature review of the literature, we identified two equally suitable types of experimental volume loading: shunt models, associated with an increased pulmonary blood flow volume and pulmonary regurgitation models, associated with a normal pulmonary blood flow volume. Based upon the present haemodynamic data, we did not find notable different hemodynamic responses. However, shunt models may be at risk for development of pulmonary vascular disease, such as pulmonary hypertension (PH) due to the increased pulmonary blood flow. If the pulmonary artery pressure increases, the RV is not only subjected to volume load, but also to pressure load, which is known to induce a different adaptation profile.[16] Other causes of RV volume overload, clinically relevant in patients with CHD, include tricuspid regurgitation (TR) and atrial septal defect (ASD). TR has been recognized as an important factor in RV failure.[31] Animal models of TR and ASD have been described, however these models were mostly used to analyse the feasibility of transcatheter repair and did not meet the inclusion criteria for this review, i.e. quantification of volume load. Nevertheless, such models may be of great interest since they allow to study the effects of unloading of the RV.[32,33]

The presented hemodynamic data in this review describe a pattern of RV adaptation to chronic experimental volume load. In line with observations in human RV adaptation to volume load, RV dilatation was consistently present in the animal models, evidenced by increased ESV/A and EDV/A. In the pressure loaded RV, as in patients with PH, RV dilatation is an early sign of failure of the initial homeometric adaptation (increased systolic function) and is accompanied by clinical deterioration.[34] In this review of the *chronically* volume loaded RV, heterometric adaptation (Starling) is the predominant mechanism involved in RV adaptation. This may be different from the initial observation by Rosenblueth et. al. from 1959.[35] In this study, the effects of RV ventricular adaptation to an *acute* rather than a *chronic* volume load were studied. This may account for the differences observed. In the volume loaded models, no other signs of overt RV failure at rest were observed, such as decreased CO or EF, although the meta-regression indicates that the initial rise in CO, predominantly present in shunt models and a

logical result of left-right shunting, declines over time. The observed decrease of RV PRSW and RV ESPVR indicates impaired systolic RV function. Intuitively, the reader may suggest a paradox in the rise in RV dP/dt max. We suggest that the rise in dP/dt max, which is a load-dependent parameter, can be explained for by the Frank-Starling mechanism: more pre-stretch increases the rate of pressure development independent from cardiac contractility. Also, RV EF did not decrease over time in response to RV volume overload, which may be expected when cardiac function is impaired.

In addition to a decline in systolic functional parameters, the systematic analysis revealed a uniform increase in RV EDP. The EDPVR is described by a curvilinear relationship, which implies that severe increase in preload is always accompanied by an increase in end-diastolic pressure. The rate at which the EDP increases is higher in "stiffer" ventricles associated with decreased diastolic function. It is unclear from the reviewed data set, whether the rise in RV EDP is in proportion to the volume load (normal diastolic function) or disproportional due to diastolic dysfunction. The latter would assume a similar profile as has been described in the pressure loaded RV.[36,37]

The lack of available data is hampering further analysis of functional adaptation. Load-independent parameters of diastolic dysfunction, i.e. the RV EDPVR or tau, a time constant of isovolumetric relaxation, were not systematically reported. Furthermore, clinical findings widely used in guidelines of human disease are scarcely reported (e.g. exercise intolerance, dyspnoea, syncope and QRS duration).[38] Borgdorff et al. have shown that systematically identifying clinical RV failure in small animal models with RV pressure overload is feasible.[37] Also, various studies have shown that exercise can be used as clinical parameter in larger animals.[39,40] We strongly recommend systematic inclusion of clinical parameters, to improve translational utility, and load independent haemodynamic analysis by pressure-volume measurements.

How can the insights regarding hemodynamic adaptation from these experimental studies be used in clinical practice? For assessment of RV volume, cardiac MRI still is the gold standard.[41] However, in the volume loaded RV, dilatation is a direct result of the changes in loading conditions rather than a first sign of deteriorating RV function in PH.[6,42] In addition, parameters assessing motion, e.g. TAPSE, are strongly affected by preload, since the velocity of shortening increases with increased pre-stretch. The function of the heart is to pump blood against a resistance, hence cardiac work is best described by parameters incorporating pressure and volume. Unfortunately, assessment of EDPVR or ESPVR is clinically often not feasible. A clinical more feasible parameter suggested to gauge RV function and its adaptation to volume load is RV stroke work (RV SW). RV SW can be measured clinically during right heart catheterisation,

and has been shown to correlate with NYHA class and outcome in specific patient groups, such as patients with heart failure and RV volume load due to TR and mixed loading conditions.[43,44] However, considering the invasiveness of catheterisation, there is increasing attention for clinically more applicable echocardiographic derivatives of SW, such as echocardiographic RV SW (estimated stroke volume times estimated RV pressure using TR velocity). Recently, echocardiographic techniques to assess RV SW have been validated also in pediatric patients with mixed loading conditions.[45] In addition, combined parameters, e.g. the product of TAPSE (motion) and dP/dt of the TR jet (pressure), correlate with outcome in adults with TR and heart failure.[46] Future prospective analyses are necessary to further validate these functional parameters in the volume loaded RV.

Multiple papers included in this review reported RV hypertrophy and fibrosis, allowing us to assess specific patterns over time. No association was demonstrated between RV weight and model type or degree and duration of volume load. This, in combination with the significant increase in RV weight in both model types, might suggest that hypertrophy is an early response mechanism, with no further progression during late remodelling. In contrast, RV fibrosis appeared to be related to prolonged volume load duration. This time-dependent development of fibrosis could be a contributing factor to the development of diastolic dysfunction. This is in line with the significant relation between fibrosis and patient age observed in clinical research.[47] Further studies investigating the role of cardiac fibrosis in the adaptation to RV volume overload are warranted to explore the therapeutic potential of anti-fibrotic treatments.

We have included 467 experimental animals in this systematic review, but unfortunately we were not able to meta-analyse any further cellular and molecular RV remodelling in response to volume load (figure 3 and table 3). The lack of systematic reports of molecular pathways involved in the process of RV adaptation may be due to underreporting of negative results. Nevertheless, using the data available, the following pattern of RV adaptation to volume overload emerges. In literature, the volume loaded left ventricle and the pressure loaded RV show an adaptation profile with hypertrophy, fibrosis, a switch to the fetal gene pathway, proliferation and inflammation.[37,48,49] Some of these components are also present in the volume loaded RV, e.g. a consistent change in β-MHC/α-MHC ratio, suggestive of a switch to the fetal gene program. However, fibrosis is less consistently reported and only emerges after several months of volume loading. Also, the ERK-pathway was de-activated, which is suggested to induce the eccentric hypertrophy and is specific for volume loading.[50] The gene-array study, performed in mice with pulmonary regurgitation, also showed a biphasic response in TGF-β activation, possibly specific for the RV, and some genes that were RV loading condition specific, e.g. SFRP2 for volume loading and BL1 and DKK3 for pressure loading.[26] Multiple serum biomarkers that are commonly reported in human heart failure, are under-studied in the included articles. For example, there is no report of soluble ST2 or galectin 3, emerging prognostic parameters of clinical heart failure in human adults.[38] Structured analysis and report of cellular and molecular pathways, that includes non-significant results, will lead to a better understanding of the pathophysiological processes involved in the development of volume overload-induced RV failure.

Study limitations

The results of this study must be considered in the context of several limitations. Substantial between-study heterogeneity was observed in the combined experiments. Heterogeneity could be due to the use of different surgical techniques and different grades of volume overload severity. Another limitation affecting heterogeneity of the analysed data is the comparison of equal volume load durations in different animal species, without taking different developmental stages and life expectancies into account. The predominant use of animals before puberty in pig models versus the predominant use of animals after puberty in rodent models could create bias, and could be a possible explanation for the high heterogeneity (table 1). Despite these limitations, we were able to obtain an overview of the currently available knowledge on the volume loaded RV in animal models.

CONCLUSION

This systematic review combined with separate meta-analyses enabled us to provide a concise overview of all available studies, to describe a pattern of ventricular adaptation and to highlight the clearly existing gaps in our knowledge of the volume loaded RV in animal models. Volume overload can be achieved by the aorto-caval shunt model and the pulmonary regurgitation model, and leads to haemodynamic adaptation characterised by systolic and diastolic RV dysfunction. Onset of RV hypertrophy due to volume load occurs early, and onset of RV fibrosis appears to occur later after volume load initiation. We recommend standardised volume load quantification and identification of involved molecular pathways in order to bridge the translational gap from bench-to-bedside in CHD.

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